Prognostic value of TARC and quantitative PET parameters in relapsed or refractory Hodgkin lymphoma patients treated with brentuximab vedotin and DHAP

HOVON Lunenburg Lymphoma Phase I/II Consortium (LLPC); Driessen, Julia; Kersten, Marie José; Visser, Lydia; van den Berg, Anke; Tonino, Sanne H.; Zijlstra, Josée M.; Lugtenburg, Pieternella J.; Morschhauser, Franck; Hutchings, Martin

Published in:
Leukemia

DOI:
10.1038/s41375-022-01717-8

Publication date:
2022

ARTICLE

LYMPHOMA

Prognostic value of TARC and quantitative PET parameters in relapsed or refractory Hodgkin lymphoma patients treated with brentuximab vedotin and DHAP

Julia Driessen[1,15], Marie José Kersten[1,15]✉, Lydia Visser[2], Anke van den Berg[2], Sanne H. Tonino[1], Josée M. Zijlstra[3], Pieternella J. Lugtenburg[4], Franck Morschhauser[5], Martin Hutchings[6], Sandy Amorim[7], Thomas Gastinne[8], Marcel Nijland[9], Gerben J. C. Zwezerijnen[10], Ronald Boellaard[10], Henrica C. W. de Vet[11], Anne I. J. Arens[12], Roelf Valkema[13], Roberto D. K. Liu[1], Esther E. E. Drees[14], Daphne de Jong[14], Wouter J. Plattel[9], Arjan Diepstra[2]✉ on behalf of the HOVON Lunenburg Lymphoma Phase I/II Consortium (LLPC)*

Risk-stratified treatment strategies have the potential to increase survival and lower toxicity in relapsed/refractory classical Hodgkin lymphoma (R/R cHL) patients. This study investigated the prognostic value of serum (s)TARC, vitamin D and lactate dehydrogenase (LDH), TARC immunohistochemistry and quantitative PET parameters in 65 R/R cHL patients who were treated with brentuximab vedotin (BV) and DHAP followed by autologous stem-cell transplantation (ASCT) within the Transplant BRaVE study (NCT02280993). At a median follow-up of 40 months, the 3-year progression free survival (PFS) was 77% (95% CI: 67–88%) and the overall survival was 95% (90–100%). Significant adverse prognostic markers for progression were weak/negative TARC staining of Hodgkin Reed-Sternberg cells in the baseline biopsy, and a high standard uptake value (SUV)mean or SUVpeak on the baseline PET scan. After one cycle of BV-DHAP, sTARC levels were strongly associated with the risk of progression using a cutoff of 500 pg/ml. On the pre-ASCT PET scan, SUVpeak was highly prognostic for progression post-ASCT. Vitamin D, LDH and metabolic tumor volume had low prognostic value. In conclusion, we established the prognostic impact of sTARC, TARC staining, and quantitative PET parameters for R/R cHL, allowing the use of these parameters in prospective risk-stratified clinical trials. Trial registration: NCT02280993.

Leukemia (2022) 36:2853–2862; https://doi.org/10.1038/s41375-022-01717-8

INTRODUCTION

Approximately 50–60% of relapsed or primary refractory classical Hodgkin lymphoma (R/R cHL) patients can be cured with standard salvage chemotherapy followed by high-dose chemotherapy (HDC) and autologous stem-cell transplantation (ASCT) [1–5]. With the advent of novel therapies for R/R cHL, optimizing baseline risk stratification and early response assessment are becoming increasingly important to guide treatment decisions [6–8].

We and others have shown that brentuximab vedotin (BV), an anti-CD30 antibody-drug conjugate, can be safely added to standard salvage chemotherapy [6, 8–14]. In the prospective, multicenter, international Phase I-II Transplant BRaVE study, we investigated the safety and efficacy of BV in combination with dexamethasone, cisplatin and high-dose cytarabine (DHAP) followed by ASCT [8]. The complete metabolic response (CMR) rate after three cycles of BV-DHAP was 100% in the Phase I part ($n = 12$) of the study and 81% in the Phase II part ($n = 55$).

To enable broader application of risk-stratified treatment, it is important to identify biomarkers that are associated with response to salvage treatment and the risk of relapse thereafter.

[1]Department of Hematology, Amsterdam UMC, University of Amsterdam, LYMMCARE, Cancer Center Amsterdam, Amsterdam, The Netherlands. [2]Department of Pathology and Medical Biology, University of Groningen, University Medical Center Groningen, Groningen, The Netherlands. [3]Department of Hematology, Amsterdam UMC, Vrije Universiteit Amsterdam, Cancer Center Amsterdam, Amsterdam, The Netherlands. [4]Department of Hematology, Erasmus MC Cancer Institute, University Medical Center, Rotterdam, The Netherlands. [5]Department of Hematology, Centre Hospitalier Universitaire, Lille, France. [6]Department of Hematology, Rigshospitalet, Copenhagen, Denmark. [7]Department of Hematology, Hopital Saint Louis, Paris, France. [8]Department of Hematology, Centre Hospitalier Universitaire, Nantes, France. [9]Department of Hematology, University of Groningen, University Medical Center Groningen, Groningen, The Netherlands. [10]Department of Radiology and Nuclear Medicine, Amsterdam UMC, Vrije Universiteit Amsterdam, Cancer Center Amsterdam, Amsterdam, The Netherlands. [11]Department of Epidemiology and Data Science, Amsterdam Public Health Research Institute, Amsterdam, The Netherlands. [12]Department of Radiology, Nuclear Medicine and Anatomy, Radboud University Medical Center, Nijmegen, The Netherlands. [13]Department of Radiology and Nuclear Medicine, Erasmus University Medical Center, Rotterdam, The Netherlands. [14]Department of Pathology, Amsterdam UMC, Vrije Universiteit Amsterdam, Cancer Center Amsterdam, Amsterdam, The Netherlands. [15]These authors contributed equally: Julia Driessen, Marie José Kersten. *A list of authors and their affiliations appears at the end of the paper. Presented in part at the European Hematology Association congress 2021 (June 9–17, 2021) and 16th International Conference on Malignant Lymphoma 2021 (June 18–22, 2021). ✉email: m.j.kersten@amsterdamumc.nl; a.diepstra@umcg.nl

Received: 17 February 2022 Revised: 20 August 2022 Accepted: 26 September 2022
Published online: 14 October 2022

Achieving a CMR, i.e., Deauville score (DS) 1–3, assessed by an ^{18}F-fluorodeoxyglucose (FDG)-positron emission tomography (PET)-computed tomography (CT) scan after salvage chemotherapy prior to ASCT, is an important predictor of progression free survival (PFS) [15–17]. The DS is determined by visual comparison of the FDG uptake in tumor localizations compared to the liver and mediastinal blood pool. However, visual assessment of DS inevitably leads to inter-observer disagreement [18]. Quantitative PET analysis leads to standardized interpretation and could provide prognostic information beyond staging and DS alone, such as metabolic tumor volume (MTV) and FDG uptake of lymphoma lesions [9, 19–21].

Besides imaging biomarkers, several blood-based and immunohistochemistry (IHC)-based markers have been investigated in newly diagnosed cHL [22–24]. Thymus and activation regulated chemokine (TARC, CCL17) is secreted by Hodgkin Reed-Sternberg (HRS) cells and can be visualized by IHC. Serum (s)TARC levels correlate with disease activity during treatment in newly diagnosed cHL [22, 24, 25]. Furthermore, serum 25-hydroxyvitamin D deficiency has been shown to correlate with poor PFS in newly diagnosed cHL patients [26, 27]. However, studies that have prospectively investigated biomarkers in the R/R setting are scarce.

Combination of blood-based, IHC-based and imaging-based biomarkers could provide prognostic information already at baseline and could complement treatment response-evaluation with visual assessment of PET-CT before ASCT. Additionally, blood-based biomarkers have the advantage that they can be assessed at multiple time points and are less invasive compared to PET-CT scans.

Here we present the 3-year follow-up results of the Transplant BRaVE study. We investigated the correlation between sTARC, tumoral TARC IHC, lactate dehydrogenase (LDH), vitamin D, quantitative PET parameters and clinical characteristics, and the prognostic value of these variables to predict progression of disease during or after BV-DHAP.

METHODS

Patients and study design

This multicenter, single-arm, Phase I-II trial (NCT02280993) enrolled adults with histologically confirmed cHL either having primary refractory disease (i.e., no complete response (CR) or progression <3 months after first-line treatment) or a first relapse after first-line chemotherapy (i.e., progression ≥3 months after CR). The complete list of inclusion and exclusion criteria has been published before [8]. Patients were treated with three cycles of BV-DHAP, followed by PET-CT response assessment. Patients with a CMR or partial metabolic response (PMR) proceeded to ASCT [8].

All patients provided written informed consent. The study protocol was approved by the Ethical Review Committee of all participating centers. The study was carried out in accordance with the principles of the Helsinki Declaration.

Serum biomarker assessment

Serum samples were centrally collected at baseline, after each cycle of BV-DHAP, after ASCT and during follow-up until 3-years post-ASCT. sTARC (ELISA, R&D systems, Minneapolis, MN, USA) and 25-hydroxyvitamin D (PromoCell, Heidelberg, Germany) levels were measured in serum by enzyme-linked immunosorbent assay, and analyzed blinded for patient outcome. LDH was not centrally analyzed but results of local laboratory assessments were collected and divided by the laboratory-specific upper limit of normal (ULN).

Tissues and immunohistochemistry

A lymph node biopsy was done at baseline, i.e., before start of BV-DHAP. For $n = 21$ patients for whom insufficient material was available for additional IHC staining, the primary diagnostic biopsy was used. Central pathology review was performed by two experienced hemato-pathologists (AD, DdJ). All cases were stained for TARC in an automated setting. Paraffin tissue sections (3 µm) were incubated with polyclonal goat-anti-human TARC antibody (1:800 R&D Systems, Minneapolis, MN) on the automated Benchmark ULTRA platform (Ultra CC1, 52 min, Roche, Ventana Medical Systems). For each TARC stain, a section of cHL tissue was applied on the same slide as an external positive control. Intensity of TARC staining (i.e., negative, weak, positive) was scored by an experienced hemato-pathologist (AD), blinded for patient outcome. Positive TARC staining was defined as cytoplasmic staining visible at a magnification of ×20 or less, weak staining was defined as cytoplasmic staining only discernable at higher magnification (×200).

PET-CT scan analysis

PET acquisition was performed according to the EANM guidelines and EARL standards in eight medical centers (Supplementary Table 1) [28, 29]. PET-CT scans were performed at baseline, prior to ASCT (4–6 weeks after the third cycle of BV-DHAP) and 6 weeks after ASCT. Central PET-CT review for response assessment according to the Lugano classification was performed by two nuclear medicine physicians (AA, RV) [8]. Discrepancies were adjudicated by a third reviewer (GJCZ).

Segmentation of baseline PET scans was performed semi-automatically using the ACCURATE tool, by automatic selection of regions with FDG uptake above a threshold of standard uptake value (SUV) ≥ 4.0 g/ml, followed by manually adding tumor regions or removing non-tumor regions with high physiological uptake if necessary, as described earlier [30, 31]. Pre-ASCT PET scans were analyzed manually if metabolic active disease was present. In patients without measurable metabolic active lesions (DS-1), SUV was set to 0 and deltaSUV to 100%. Regarding extranodal and splenic lesions, only focal lesions were included. PET segmentation was performed by JD under supervision of a nuclear medicine physician (GJCZ). The following quantitative features were extracted at the patient-level: SUVmean, SUVpeak, total MTV, total lesion glycolysis (TLG; i.e., MTV multiplied by SUVmean), and number of lesions [19, 32]. Because of the multicenter aspect of this study and the use of different PET scanners, only PET parameters that are not too sensitive to technical variations were used, such as SUVpeak (i.e., the average SUV of the 1 ml with the highest FDG uptake) instead of SUVmax (which represents only the highest single voxel). Additionally, as the SUVmean of the liver is used as a standard quality parameter to compare PET scans and is also the reference for a DS-3, we normalized the SUV for the liver SUVmean and used the tumor-to-liver ratio (TLR) [28, 33–35]. The liver SUVmean was estimated on a 3 ml sphere in the right upper lobe of the liver. In addition, we calculated the tumor-ratio for the mediastinal blood-pool (MBP), which is the reference for a DS-2.

Endpoints

The efficacy and safety endpoints of the Transplant BRaVE phase I–II study have been reported earlier [8]. The endpoint of the clinical follow-up analysis is the 3-year PFS and overall survival (OS). PFS was defined as time from study entry until progressive disease or death, whichever came first. OS was defined as time from study entry until death from any cause. The primary endpoint for biomarker analysis is the 3-year freedom from progression (FFP), defined as time from study entry until progressive disease, and patients who died without progression were censored at the time of death. This provides a more biologically meaningful analysis of the correlation between the biomarkers and disease activity.

Statistical analysis

The Kaplan–Meier method and log-rank test were used to analyze univariable associations with PFS and OS and a Cox proportional hazards regression was performed. Biomarker values were compared for patients who showed progressive disease during or after BV-DHAP vs. patients in remission using the Wilcoxon rank sum test for non-parametric data. Correlations between biomarkers were assessed using Spearman's Rank correlation coefficients. The prognostic value of biomarkers for FFP was assessed by calculating the area under the curve (AUC) of the receiver operating characteristics curve and log-rank survival analysis. Added prognostic value of combining two biomarkers was assessed using logistic regression and Wald test. Pre-specified cutoffs were used to calculate sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV). The pre-specified cutoff for response-assessment with sTARC was 1000 pg/ml, which is based on a study in newly diagnosed cHL and levels in healthy controls [22]. Patients who had sTARC-baseline levels below the cutoff were excluded from subsequent analysis. The cutoff for sufficient vitamin D levels was 50 ng/ml [26, 27]. The cutoff for baseline $TLR_{SUVmean}$ and $TLR_{SUVpeak}$ was 3.0, aligning with a DS-5 (uptake markedly higher than the

liver), and for pre-ASCT $TLR_{SUVmean}$ and $TLR_{SUVpeak}$, a cutoff of 1.0 was used, aligning with DS-3 [35]. Missing values of sTARC-1 were replaced by sTARC levels after cycle 2 or 3 for eight respective patients. Missing values of sTARC-3 were replaced by sTARC levels after cycle 1 or 2 for six respective patients. Sensitivity analyses were performed with and without replacement for missing values. For other variables, no missing values were replaced. Clinical data were collected using OpenClinica version 3.6 [36], and the statistical analysis was performed in R software version 4.0.

RESULTS

Patients and treatment

Between May 2014 and July 2017, 67 patients with R/R cHL were enrolled in the Transplant BRaVE study ($n = 12$ in Phase I, and $n = 55$ in Phase II) (Table 1). Two patients withdrew consent after one cycle of BV-DHAP due to psychological issues and were excluded from further analyses. Seven patients were reclassified as non-Hodgkin lymphoma (e.g., peripheral T-cell lymphoma) according to central pathology review and were excluded from biomarker analyses, but not from evaluation of clinical endpoints per intention to treat [8].

Table 1. Baseline demographics and disease characteristics.

	cHL ($N = 58$)	Other ($N = 7$)	Total ($N = 65$)
Age at relapse (years)			
Median (Min–Max)	29 (19–64)	30 (20–63)	29 (19–64)
Disease status			
Refractory	26 (45%)	4 (57%)	30 (46%)
Relapse <1 year	16 (28%)	–	16 (25%)
Relapse ≥1 year	16 (28%)	3 (43%)	19 (29%)
Stage at relapse			
I/II	27 (47%)	3 (43%)	30 (46%)
III/IV	31 (53%)	4 (57%)	35 (54%)
B symptoms			
Yes	18 (31%)	5 (71%)	23 (35%)
Extranodal disease			
Yes	25 (43%)	1 (14%)	26 (40%)
Splenic focal lesions			
Yes	8 (14%)	1 (14%)	9 (14%)
Morphological subtype			
NS	38 (66%)	–	38 (58%)
MC	13 (22%)	–	13 (20%)
NOS	7 (12%)	–	7 (11%)
AITL	–	1 (14%)	1 (2%)
IA-B-LPD	–	1 (14%)	1 (2%)
PTCL	–	5 (71%)	5 (7%)
EBV positive			
Yes	7 (13%)	7 (100%)	14 (23%)
Missing	3	–	3
TARC staining			
Positive	43 (86%)	4 (57%)	47 (83%)
Weak	4 (8%)	2 (29%)	6 (11%)
Negative	3 (6%)	1 (14%)	4 (7%)
Missing	8	–	8
Events			
Progression	11 (19%)	2 (29%)	13 (20%)
Death	2 (3%)	1 (14%)	3 (5%)

cHL classical Hodgkin lymphoma, *N* number of patients, *NS* nodular sclerosis, *MC* mixed cellularity, *NOS* not otherwise specified, *AITL* angioimmunoblastic T-cell lymphoma, *IA-B-LPD* immunodeficiency-associated B-lymphoproliferative disorder, *PTCL* peripheral T-cell lymphoma not otherwise specified, *EBV* Epstein-Barr virus, *TARC* thymus and activation regulated chemokine.

Long-term follow-up results

The median follow-up time was 40 months (range 23–65) in patients still alive at time of the analysis. The 3-year PFS by intention-to-treat for all 65 patients was 77% (95% confidence interval; CI: 67–88%) and the OS was 95% (95% CI: 90–100%) (Fig. 1A). In total, three patients died ($n = 2$ cHL, $n = 1$ peripheral T-cell lymphoma), all without signs of progressive disease [8]. The 3-year FFP in patients with confirmed cHL diagnosis who were included in the biomarker analyses was 82% (95% CI: 73–93) (Fig. 1B).

Serum TARC

The median sTARC-baseline level was 4885 pg/ml (range 282–120,654) and significantly decreased to 384 pg/ml (113–28,448) after cycle 1 ($p < 0.0001$) (Fig. 2A). sTARC-baseline did not differ significantly between patients who relapsed and patients in remission after BV-DHAP (median 5204 vs. 3600 pg/ml; $p = 0.9$), and was not prognostic for FFP (AUC 0.49) (Supplementary Table 2). The percentual drop in sTARC levels after cycle 1 (deltaTARC-1) was larger in patients with favorable outcomes but showed only moderate prognostic value (AUC 0.66) (Fig. 2B and Supplementary Table 2).

sTARC after cycle 1 (sTARC-1) was significantly higher in patients who relapsed during or after BV-DHAP in comparison to patients in remission (median 889 vs. 338 pg/ml; $p = 0.008$) (Fig. 2C). This was also the case for sTARC after cycle 2 (sTARC-2) ($p = 0.017$) and sTARC after cycle 3 (sTARC-3) ($p = 0.009$). sTARC-1 had strong prognostic value for FFP (AUC 0.76) (Supplementary Table 2). Sensitivity analysis showed no differences in prognostic value of sTARC-1 when patients with missing values were excluded (Supplementary Tables 2 and 3).

A predefined cutoff of 1000 pg/ml was used based on levels in healthy controls and use in a clinical setting to have high specificity for newly diagnosed cHL patients [22]. However, compared to sTARC levels described in newly diagnosed cHL patients, sTARC-baseline levels were much lower in our R/R cHL cohort (median serum TARC 28,013 vs. 4885 pg/ml, respectively) [22], and $n = 14$ patients had sTARC-baseline levels <1000 pg/ml. Therefore, we decided to use a lower sTARC cutoff of 500 pg/ml for response evaluation in addition to the pre-specified cutoff of 1000 pg/ml. The pre-specified cutoff of 1000 pg/ml could significantly discriminate patients with a favorable FFP (3-year FFP 90% vs. 55%; $p = 0.01$) (Fig. 2D). Only four patients had sTARC-baseline levels <500 pg/ml. The cutoff of 500 pg/ml for sTARC-1 provided strong significant discrimination between patients with favorable and unfavorable FFP (3-year FFP 96% vs. 64%; $p = 0.003$) (Fig. 2E). Additionally, when excluding the four patients with a sTARC-baseline <500 pg/ml ($n = 4$), the AUC of sTARC-1 increased from 0.76 to 0.81 (Supplementary Table 2).

sTARC-3 levels were higher in patients with a PMR (DS 4–5) or progressive disease on the pre-ASCT PET scan, but this was not statistically significant (Supplementary Fig. 1A).

For patients with progressive disease during follow-up, sTARC levels at time of progression were ≥500 pg/ml in 7/9 patients (78%) (Supplementary Fig. 1B, C). Taking sTARC levels of all time points of patients with a CMR during follow-up ($n = 278$ time points) compared to sTARC levels from patients at time of progression ($n = 9$), sTARC showed a PPV of only 8% for detecting progressive disease and a NPV of 99% for excluding progressive disease using a cutoff of 500 pg/ml (Supplementary Fig. 1C).

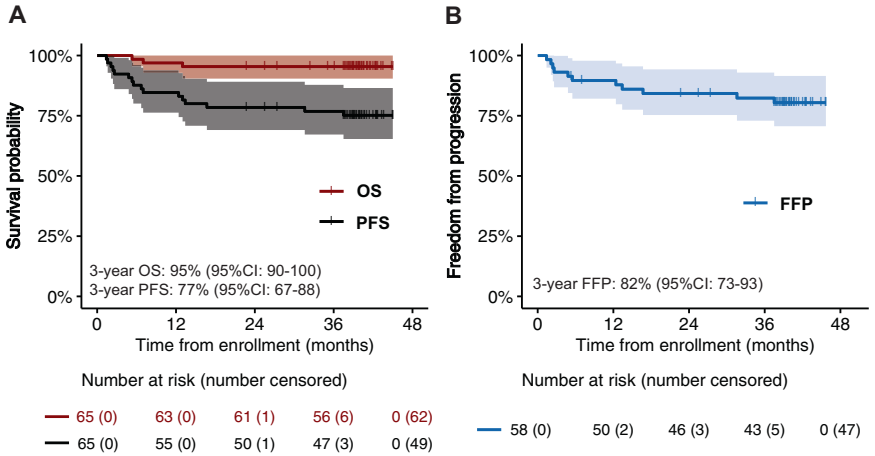

Fig. 1 Progression free survival, overall survival, and freedom from progression in patients with R/R cHL. A Three-year progression free survival (PFS) and overall survival (OS) in all patients enrolled in the study. **B** Freedom from progression (FFP) in all patients with confirmed classical Hodgkin lymphoma diagnosis during central pathology who were included in the biomarker analyses. Patients who had a diagnosis other than cHL on central pathology review ($n = 7$) were excluded. Patients who died without progression ($n = 2$) were censored at time of death. OS overall survival, PFS progression free survival, FFP freedom from progression.

25-hydroxyvitamin D and LDH

Baseline serum 25-hydroxyvitamin D levels indicated deficiency (<30 ng/ml) in four patients (7%) and insufficiency (30–50 ng/ml) in 16 patients (29%). Patients with primary refractory disease had lower vitamin D levels compared to relapsed patients ($p = 0.018$). Vitamin D levels as a continuous variable had low prognostic value for FFP (AUC 0.57), and there were no significant differences in vitamin D levels between patients with or without progression ($p = 0.52$) or patients with a CMR vs. PMR ($p = 0.92$) or progression ($p = 0.62$) on the pre-ASCT PET scan (Supplementary Fig. 2 and Supplementary Tables 2 and 3).

LDH was elevated (≥1 ULN) at baseline in 13 patients, but these patients did not show a higher incidence of progression ($p = 0.5$). LDH levels were not significantly higher in patients who progressed compared to patients without progression ($p = 0.13$) and there were no differences in pre-ASCT LDH levels for patients with a CMR vs. PMR ($p = 0.16$) or progression ($p = 0.54$) on the pre-ASCT PET scan. LDH significantly increased during BV-DHAP treatment, and after ASCT decreased to normal levels for most patients, probably coinciding with administration of granulocyte colony-stimulating factor (Supplementary Fig. 3).

TARC immunohistochemistry

In total, 50 out of 58 confirmed cHL patients had a lymph node biopsy available for additional IHC staining. All patients stained positive for CD30 in HRS cells. Forty-three patients (86%) stained positive for TARC in the HRS cells (Fig. 3A). Patients with negative or weak TARC staining ($n = 7$) showed significantly lower sTARC-baseline levels compared to patients with positive TARC staining in the HRS cells (median 608 vs. 3701 pg/ml, respectively; $p = 0.04$) (Fig. 3B). More importantly, these patients showed a significant lower 3-year FFP compared to patients with positive TARC staining in the HRS cells (3-year FFP 89% vs. 48%; $p = 0.0004$) (Fig. 3C).

Quantitative PET scan analysis

Baseline $TLR_{SUVmean}$ and $TLR_{SUVpeak}$, were higher in patients who progressed during or after BV-DHAP compared to patients in remission ($p < 0.001$ and $p = 0.04$, respectively) (Fig. 4A, B). Similar differences were observed for $TLR_{SUVmean}$ and $TLR_{SUVpeak}$ after three cycles of BV-DHAP prior to ASCT ($p < 0.001$ and $p < 0.001$) and after ASCT ($p = 0.01$ and $p = 0.03$), respectively (Fig. 4A, B). Patients who progressed during or after treatment also showed a lower $deltaTLR_{SUVmean}$ and $deltaTLR_{SUVpeak}$ (Fig. 4C, D). Only one patient who relapsed after 3 years had a $deltaTLR_{SUVmean}$ of −100%. Prognostic value as estimated by AUC was low for MTV and TLG (0.47 and 0.54, respectively), and highest in baseline $TLR_{SUVmean}$ and $TLR_{SUVpeak}$ (AUC 0.85 and 0.70, respectively) (Supplementary Table 2). The predefined cutoffs of $TLR_{SUVmean}$ and $TLR_{SUVpeak}$ of ≥3.0 at baseline could significantly discriminate patients in low and high-risk groups for FFP ($p < 0.0001$ and $p = 0.027$, respectively), with an NPV of 94% and a PPV of 50% for $TLR_{SUVmean}$, and an NPV of 100% and a PPV of 28% for $TLR_{SUVpeak}$ (Fig. 4E, F and Supplementary Table 3). Prognostic value of $TLR_{SUVmean}$ (AUC 0.73) and $TLR_{SUVpeak}$ (AUC 0.76) at the pre-ASCT PET-CT was higher compared to visual DS (AUC 0.69), but was comparable in terms of NPV/PPV when using a cutoff of TLR ≥ 1.0 (Fig. 4G–I and Supplementary Tables 2 and 3). Results for $MBP_{SUVmean}$ and $MBP_{SUVpeak}$ showed similar results compared to $TLR_{SUVmean}$ and $TLR_{SUVpeak}$ and are summarized in Supplementary Tables 2 and 3.

Correlations and combinations of biomarkers

There was a significant moderate to high correlation between sTARC-baseline and several PET parameters, such as MTV ($r = 0.54$) and $TLR_{SUVpeak}$ ($r = 0.4$) (Fig. 5A). Serum vitamin D levels did not show any correlation with PET parameters. LDH showed moderate correlation with $TLR_{SUVpeak}$ ($r = 0.36$). Hemoglobin showed a negative correlation with MTV ($r = -0.26$) and number of lesions ($r = -0.31$) (Supplementary Fig. 4). Patients with B symptoms had significantly higher baseline MTV ($p = 0.014$), $TLR_{SUVpeak}$ ($p = 0.006$) and LDH ($p = 0.013$), but there were no differences in sTARC-baseline levels ($p = 0.95$) (Supplementary Fig. 5).

An explorative multivariable analysis showed an increased AUC for the combinations of sTARC-1 and baseline $TLR_{SUVmean}$ (AUC 0.85) or $TLR_{SUVpeak}$ (AUC 0.77), with both variables showing an independent prognostic value ($p < 0.05$) (Supplementary Fig. 6). Patients who had both a high baseline $TLR_{SUVmean}$ (≥3.0), and high sTARC-1 (≥500 pg/ml) ($n = 13$) showed significantly lower 3-year FFP compared to patients who had either low $TLR_{SUVmean}$ or low sTARC-1 (35% vs. 95%; $p < 0.0001$), with an NPV of 95% and a PPV of 67% (Fig. 5B and Supplementary Table 4). Similarly, patients who showed both a high pre-ASCT $TLR_{SUVpeak}$ (≥1.0) and high sTARC-3 (≥500 pg/ml) ($n = 4$) showed the highest risk of progression with a 3-year FFP of 0% vs. 95% for other patients ($p < 0.0001$), with an NPV of 95% and a PPV of 100% (Fig. 5C and Supplementary Table 4).

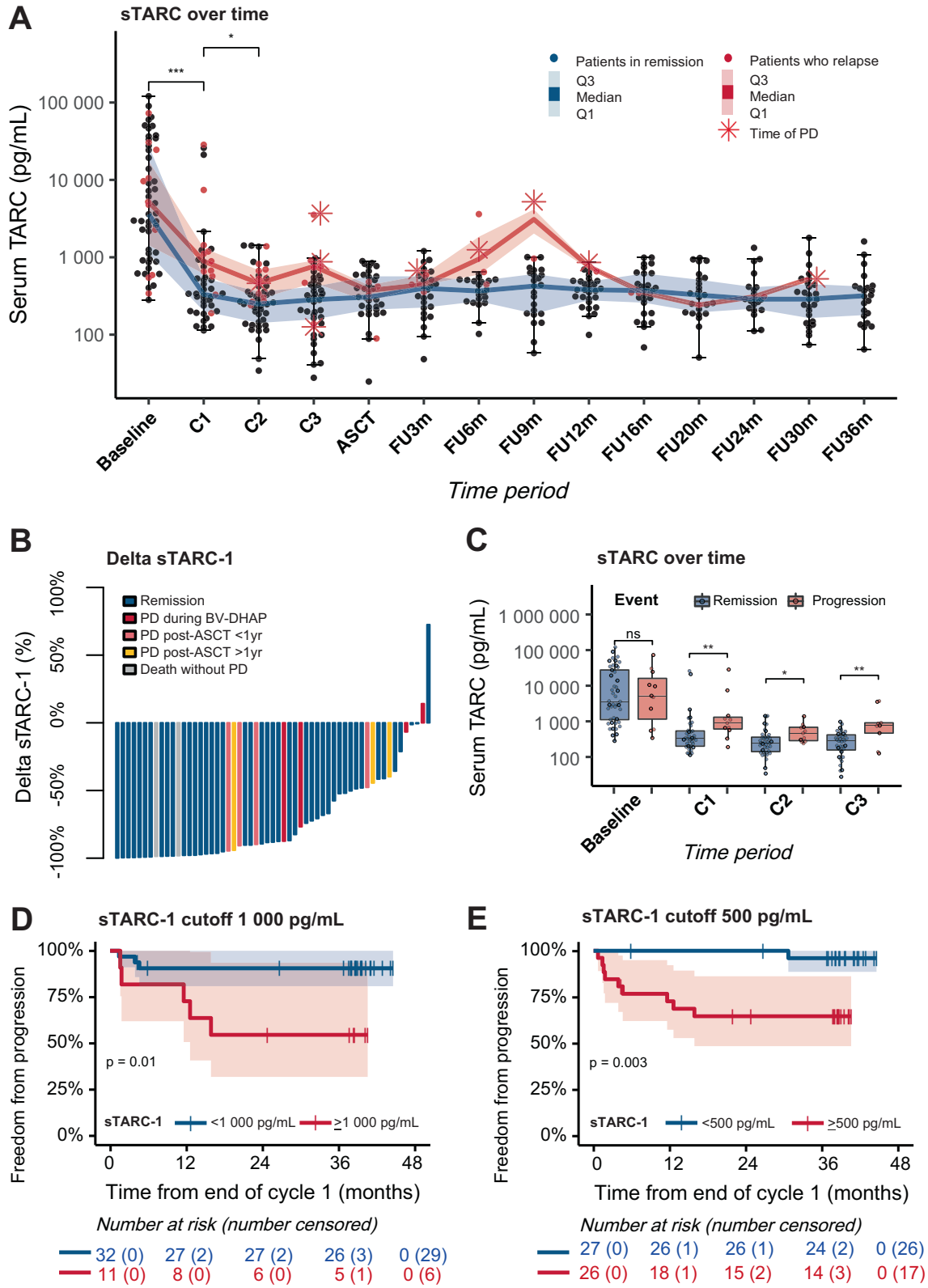

Fig. 2 Serum TARC levels during BV-DHAP treatment in R/R cHL. A sTARC at baseline, during treatment and follow-up, stratified for patients in remission (blue) and patients who developed progressive disease during or after BV-DHAP treatment (red). Dots represent sTARC values at indicated time points. The red stars represent the specific time point of progressive disease of an individual patient. Lines represent median values, bands indicate interquartile ranges (Q1–Q3). **B** Delta sTARC (%) after cycle 1 stratified for patients with and without progression during or after treatment. **C** sTARC stratified for patients in remission vs. progression on BV-DHAP at baseline, and after each cycle of BV-DHAP. Freedom from progression (FFP) Kaplan–Meier analysis for sTARC levels after cycle 1 with a cutoff of 1000 (**D**) and 500 (**E**) pg/ml. *$p < 0.05$; **$p < 0.01$; ***$p < 0.001$; ns not significant. s serum, TARC thymus and activation regulated chemokine, C cycle, ASCT autologous stem-cell transplantation, FU follow-up, m months, Q1 first interquartile, Q3 third interquartile, PD progressive disease, Tx treatment, yr year.

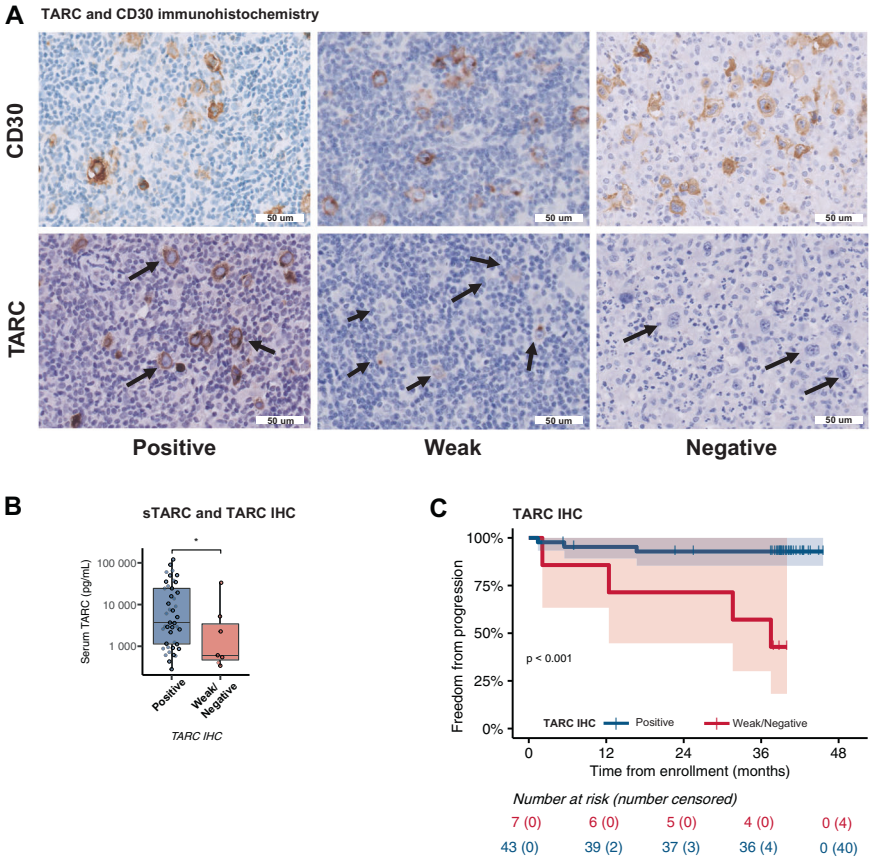

Fig. 3 TARC immunohistochemistry of lymph node biopsies at baseline. A TARC and CD30 immunohistochemistry in the lymph node biopsy of a patient with positive TARC staining (left), weak TARC staining (middle), and a patient with negative TARC staining (right). Arrows indicate HRS cells with positive, weak or negative TARC staining. Images were captured at ×20 magnification. **B** Baseline sTARC levels for patients with weak or negative TARC staining in the HRS cells vs. patients with positive TARC staining. **C** Freedom from progression (FFP) Kaplan–Meier analysis for patients with negative/weak TARC staining compared to positive TARC staining in the HRS cells of baseline lymph node biopsies. *$p < 0.05$; **$p < 0.01$; ***$p < 0.001$; ns not significant. TARC thymus and activation regulated chemokine.

DISCUSSION

This long-term follow-up analysis of the Transplant BRaVE study, investigating the addition of BV to DHAP followed by ASCT in patients with R/R cHL, showed a high 3-year PFS and OS. The 3-year PFS of 77% is higher compared to historical controls in patients treated with DHAP only, or other chemotherapy-based salvage regimens, in which the 2–5 year PFS is ~50–60% [15, 37–39]. Because the vast majority of progressions occur within 2–3 years of follow-up, the PFS rate after 3 years is a good surrogate for cure [3]. OS appears to be higher than previously reported, but this may be partially explained by the use of other novel therapies in patients who failed BV-DHAP/ASCT (e.g., checkpoint inhibition) [40]. We show that sTARC-1 is a strong prognostic biomarker in R/R cHL. Additionally, we identified several baseline and response-assessment biomarkers with prognostic value for 3-year FFP, including TARC IHC of HRS cells in tissue, and baseline and pre-ASCT $TLR_{SUVmean}$ and $TLR_{SUVpeak}$.

Strong points of this study are the prospective design regarding sample and data collection, the use of predefined cutoffs for sTARC based on results in healthy controls, and cutoffs for the SUV ratio to the liver SUVmean based on response-assessment by DS [22, 35]. The latter also justifies the comparison of quantitative PET parameters over time and between patients in different hospitals [22, 34, 35]. Limitations of this study are the small sample size and low number of events, which precluded cross-validation of results. Therefore, the possibly more optimal cutoff for sTARC-1 of 500 pg/ml instead of 1000 pg/ml, TARC IHC in tissue, and prognostic PET parameters need validation in other R/R cHL cohorts.

The high prognostic value of baseline and pre-ASCT $TLR_{SUVmean}$ and $TLR_{SUVpeak}$ warrants further exploration of using quantitative PET parameters for response-assessment and baseline risk stratification in R/R cHL. This can easily be implemented in clinical practice since the PET scan is performed at baseline and pre-ASCT as standard of care. Regarding to baseline PET measurements, $TLR_{SUVmean}$ showed higher prognostic value compared to $TLR_{SUVpeak}$, while $TLR_{SUVmean}$ and $TLR_{SUVpeak}$ had comparable prognostic value in the pre-ASCT setting. Considering low metabolic residual disease on the pre-ASCT PET in most patients, SUVpeak of the most FDG-avid lesion is easier to measure compared to patient-level SUVmean which requires segmentation of the total MTV.

In newly diagnosed cHL patients, sTARC is a strong, early response marker [22]. We showed that in R/R patients, sTARC can be used as a response marker already after one cycle of BV-DHAP. Moreover, the combination of sTARC-1 and $TLR_{SUVmean}$ or $TLR_{SUVpeak}$ provides complementary prognostic information, and identified the majority of patients who progressed. Therefore, patients having both a high baseline (≥3.0) $TLR_{SUVmean}$ and high sTARC-1 (≥500 pg/ml), or a high pre-ASCT $TLR_{SUVpeak}$ (≥1.0) and high sTARC-3 (≥500), could be regarded as high-risk for progressive disease. These patients potentially would benefit from additional treatment, for example with post-ASCT consolidation or maintenance treatment with BV or checkpoint inhibitors, which should be studied in prospective clinical trials [10, 41, 42].

Despite the strong prognostic value of sTARC, it still shows a low PPV for detecting progressive disease during follow-up [22, 23].

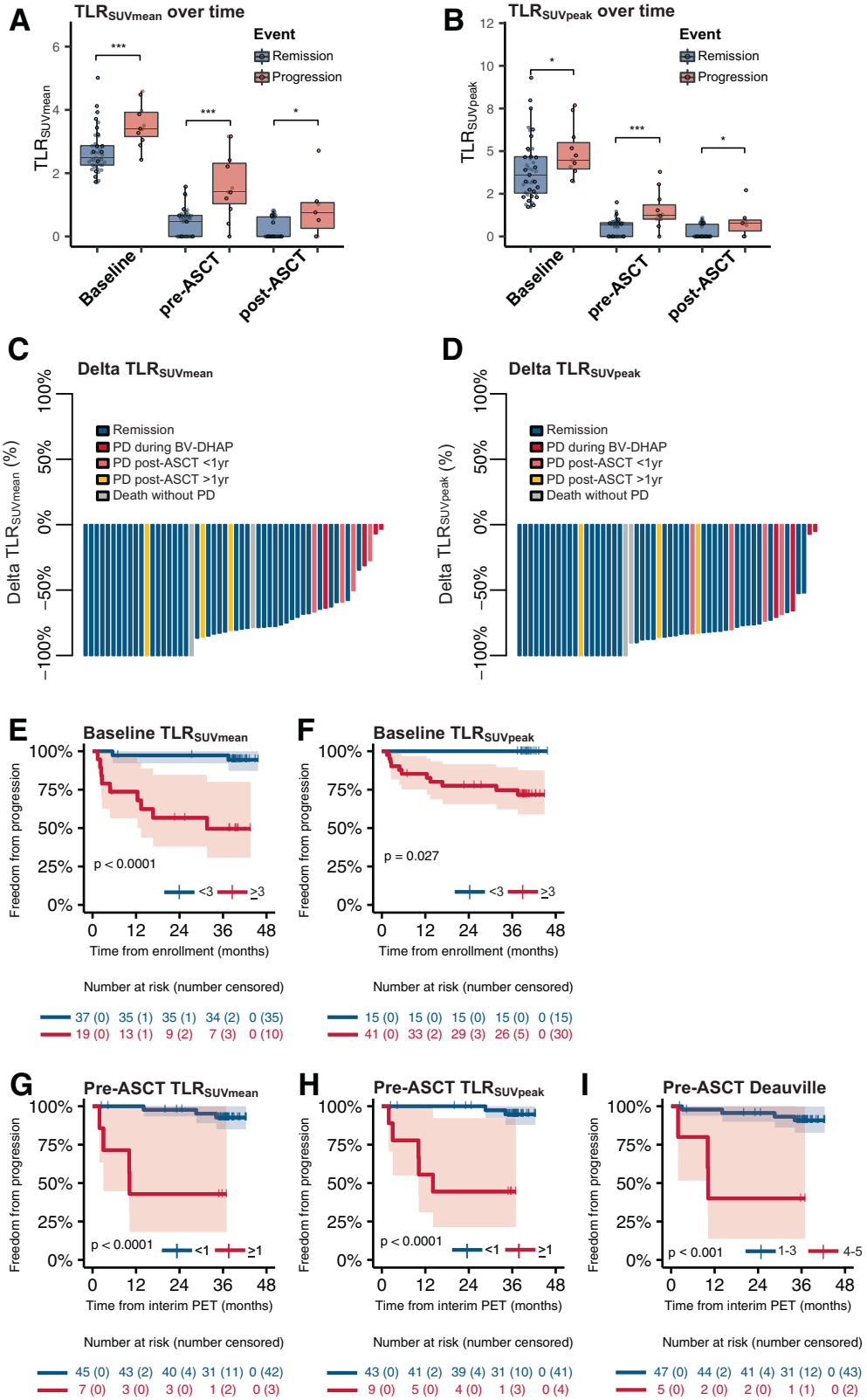

Fig. 4 Quantitative baseline and pre-ASCT PET parameters. A $TLR_{SUVmean}$ and **B** $TLR_{SUVpeak}$, on the PET-CT at baseline, pre-ASCT and post-ASCT, stratified for patients who are in remission after 3-years of follow-up compared to patients who had progressive disease during or after treatment. **C** $DeltaTLR_{SUVmean}$ and **D** $deltaTLR_{SUVpeak}$ pre-ASCT, ranked by $deltaTLR_{SUV}$. **E** Kaplan–Meier FFP analysis for $TLR_{SUVmean}$ at baseline. **F** Kaplan–Meier FFP analysis for $TLR_{SUVpeak}$ at baseline. **G** Kaplan–Meier FFP analysis for pre-ASCT $TLR_{SUVmean}$. **H** Kaplan–Meier FFP analysis for pre-ASCT $TLR_{SUVpeak}$. **I** Kaplan-Meier FFP analysis for pre-ASCT Deauville scores with a cutoff of DS1-3 for CMR. All SUVs represent ratios of tumor-to-liver SUV, corrected for the liver SUVmean. *$p < 0.05$; **$p < 0.01$; ***$p < 0.001$; ns not significant. TLR tumor-to-liver ratio, SUV standard uptake value, ASCT autologous stem-cell transplantation, Tx treatment, yr year, DS Deauville score, FFP freedom from progression.

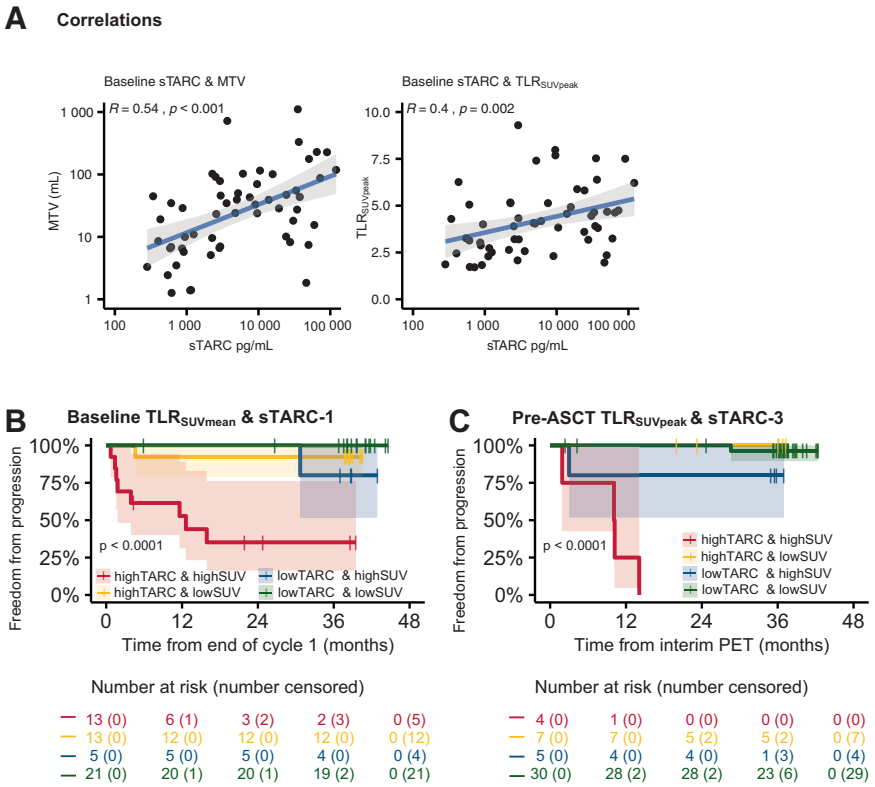

Fig. 5 Correlations and combinations of several biomarkers. A Spearman's rank correlations of sTARC and MTV and $TLR_{SUVpeak}$ at baseline. **B** Kaplan–Meier analysis for FFP stratified for patients with high (≥3.0) vs. low (<3.0) $TLR_{SUVmean}$ at baseline and high (≥500 pg/ml) vs. low (<500 pg/ml) sTARC-1. **C** Kaplan–Meier analysis for post-ASCT FFP stratified for patients with high (≥500 pg/ml) vs. low (<500 pg/ml) pre-ASCT sTARC and high (≥1.0) vs. low (<1.0) pre-ASCT $TLR_{SUVpeak}$. *$p < 0.05$; **$p < 0.01$; ***$p < 0.001$; ns not significant. MTV metabolic tumor volume, sTARC serum thymus and activation regulated chemokine, TLR tumor liver ratio, SUV standard uptake value, sTARC-1 sTARC after cycle 1, sTARC-3 sTARC after cycle 3, ASCT autologous stem-cell transplantation.

Therefore, sTARC may be less suitable as follow-up marker in the R/R setting. It should be noted, however, that in a small study in patients who relapsed after allogeneic transplantation, all seven patients showed sTARC levels ≥1000 pg/ml at time of progression [43]. In our cohort, sTARC-baseline levels (median 4885 pg/ml) were increased compared to healthy controls (median 118 pg/ml), but less pronounced as compared to earlier published data of newly diagnosed cHL patients (median 28,013 pg/ml) [22]. This may in part be explained by the generally lower tumor load as per MTV in the R/R setting, which correlates with lower sTARC-baseline levels [23]. Therefore we used a lower cutoff of 500 pg/ml in addition to the pre-specified cutoff of 1000 pg/ml. However, this cutoff should be validated in other prospective studies in R/R cHL patients.

Despite the prognostic value in newly diagnosed patients, vitamin D levels did not show prognostic value in our cohort [26, 27]. We found that patients with primary refractory disease had lower vitamin D levels compared to relapsed patients, which could be explained by either a shorter time to first-line treatment and hospital admission and thus lack of sun exposure in primary refractory patients, or by an increased chance of being primary refractory after first-line treatment when patients already have low vitamin D levels. This can however not be concluded from our data and should be investigated in other prospective studies.

Analysis of cell-free DNA (cfDNA) is emerging as a measure for minimal residual disease. It was recently shown that individual mutational fingerprints correlate with response in newly diagnosed cHL patients [44]. However, cfDNA is an expensive technique and requires complex analysis as compared to measuring sTARC. Combination of sTARC and cfDNA might provide additional prognostic information and studies combining these biomarkers are needed.

This is the first study to show prognostic value of TARC expression in HRS cells as measured by IHC in tissue biopsy samples. The mechanism behind this association is not clear and may be related to characteristics of the HRS cells, or the influence of TARC on the composition of the tumor micro-environment [45].

With the advent of highly effective novel therapies such as BV and checkpoint inhibition, one of the next goals for clinical trials is to investigate whether some R/R cHL patients can possibly be cured without HDCT/ASCT. Risk-stratified and PET-adapted prospective studies could help to identify patients who have low-risk of relapse and can be cured with salvage therapy alone, and on the other hand, identify patients who are chemotherapy-refractory early, so they can receive alternative therapies such as checkpoint inhibition.

In conclusion, we have shown a high 3-year PFS and OS with three cycles of BV-DHAP followed by ASCT in R/R cHL. sTARC can be used as an early response marker already after one cycle of BV-DHAP, and combination with $TLR_{SUVmean}$ and $TLR_{SUVpeak}$ at baseline and pre-ASCT provides strong prognostic information which can help to identify patients with high risk of progression early in the treatment course.

DATA AVAILABILITY

De-identified data will be shared with other researchers upon reasonable request to the corresponding authors (m.j.kersten@amsterdamumc.nl or a.diepstra@umcg.nl). The sharing will require a detailed proposal to the study investigators, and a data transfer agreement must be signed.

REFERENCES

1. Schmitz N, Pfistner B, Sextro M, Sieber M, Carella AM, Haenel M, et al. Aggressive conventional chemotherapy compared with high-dose chemotherapy with autologous haemopoietic stem-cell transplantation for relapsed chemosensitive Hodgkin's disease: a randomised trial. Lancet. 2002;359:2065–71.
2. Linch DC, Winfield D, Goldstone AH, Moir D, Hancock B, McMillan A, et al. Dose intensification with autologous bone-marrow transplantation in relapsed and resistant Hodgkin's disease: results of a BNLI randomised trial. Lancet. 1993;341:1051–4.
3. Majhail NS, Weisdorf DJ, Defor TE, Miller JS, McGlave PB, Slungaard A, et al. Long-term results of autologous stem cell transplantation for primary refractory or relapsed Hodgkin's lymphoma. Biol Blood Marrow Transpl. 2006;12:1065–72.
4. Josting A, Rudolph C, Mapara M, Glossmann JP, Sieniawski M, Sieber M, et al. Cologne high-dose sequential chemotherapy in relapsed and refractory Hodgkin lymphoma: results of a large multicenter study of the German Hodgkin Lymphoma Study Group (GHSG). Ann Oncol. 2005;16:116–23.
5. Moskowitz CH, Nimer SD, Zelenetz AD, Trippett T, Hedrick EE, Filippa DA, et al. A 2-step comprehensive high-dose chemoradiotherapy second-line program for relapsed and refractory Hodgkin disease: analysis by intent tot treat and development of a prognostic model. Blood. 2001;97:616–23.
6. Advani R, Moskowitz AJ, Bartlett NL, Vose J, Ramchandren R, Feldman T, et al. Brentuximab vedotin in combination with nivolumab in relapsed or refractory Hodgkin lymphoma: 3-year study results. Blood. 2021;138:427–38.
7. Moskowitz AJ, Shah G, Schöder H, Ganesan N, Drill E, Hancock H, et al. Phase II trial of pembrolizumab plus gemcitabine, vinorelbine, and liposomal doxorubicin as second-line therapy for relapsed or refractory classical Hodgkin lymphoma. J Clin Oncol. 2021;39:3109–17.
8. Kersten MJ, Driessen J, Zijlstra JM, Plattel WJ, Morschhauser F, Lugtenburg PJ, et al. Combining brentuximab vedotin with dexamethasone, high-dose cytarabine and cisplatin as salvage treatment in relapsed or refractory Hodgkin lymphoma: the phase II HOVON/LLPC Transplant BRaVE study. Haematologica. 2021;106:1129–37.
9. Moskowitz AJ, Schoder H, Gavane S, Thoren KL, Fleisher M, Yahalom J, et al. Prognostic significance of baseline metabolic tumor volume in relapsed and refractory Hodgkin lymphoma. Blood. 2017;130:2196–203.
10. Moskowitz CH, Nademanee A, Masszi T, Agura E, Holowiecki J, Abidi MH, et al. Brentuximab vedotin as consolidation therapy after autologous stem-cell transplantation in patients with Hodgkin's lymphoma at risk of relapse or progression (AETHERA): a randomised, double-blind, placebo-controlled, phase 3 trial. Lancet. 2015;385:1853–62.
11. Herrera AF, Palmer J, Martin P, Armenian S, Tsai NC, Kennedy N, et al. Autologous stem-cell transplantation after second-line brentuximab vedotin in relapsed or refractory Hodgkin lymphoma. Ann Oncol. 2018;29:724–30.
12. Garcia-Sanz R, Sureda A, de la Cruz F, Canales M, Gonzalez AP, Pinana JL, et al. Brentuximab vedotin and ESHAP is highly effective as second-line therapy for Hodgkin lymphoma patients (long-term results of a trial by the Spanish GELTAMO Group). Ann Oncol. 2019;30:612–20.
13. LaCasce AS, Bociek RG, Sawas A, Caimi P, Agura E, Matous J, et al. Brentuximab vedotin plus bendamustine: a highly active first salvage regimen for relapsed or refractory Hodgkin lymphoma. Blood. 2018;132:40–8.
14. Broccoli A, Argnani L, Botto B, Corradini P, Pinto A, Re A, et al. First salvage treatment with bendamustine and brentuximab vedotin in Hodgkin lymphoma: a phase 2 study of the Fondazione Italiana Linfomi. Blood Cancer J. 2019;9:100.
15. Moskowitz CH, Matasar MJ, Zelenetz AD, Nimer SD, Gerecitano J, Hamlin P, et al. Normalization of pre-ASCT, FDG-PET imaging with second-line, non-cross-resistant, chemotherapy programs improves event-free survival in patients with Hodgkin lymphoma. Blood. 2012;119:1665–70.
16. Moskowitz CH, Yahalom J, Zelenetz AD, Zhang Z, Filippa D, Teruya-Feldstein J, et al. High-dose chemo-radiotherapy for relapsed or refractory Hodgkin lymphoma and the significance of pre-transplant functional imaging. Br J Haematol. 2010;148:890–7.
17. Devillier R, Coso D, Castagna L, Brenot Rossi I, Anastasia A, Chiti A, et al. Positron emission tomography response at the time of autologous stem cell transplantation predicts outcome of patients with relapsed and/or refractory Hodgkin's lymphoma responding to prior salvage therapy. Haematologica. 2012;97:1073–9.
18. Burggraaff CN, Cornelisse AC, Hoekstra OS, Lugtenburg PJ, De Keizer B, Arens AIJ, et al. Interobserver agreement of interim and end-of-treatment (18)F-FDG PET/CT in diffuse large B-cell lymphoma: impact on clinical practice and trials. J Nucl Med. 2018;59:1831–6.
19. Cottereau AS, Versari A, Loft A, Casasnovas O, Bellei M, Ricci R, et al. Prognostic value of baseline metabolic tumor volume in early-stage Hodgkin lymphoma in the standard arm of the H10 trial. Blood. 2018;131:1456–63.
20. Song MK, Chung JS, Lee JJ, Jeong SY, Lee SM, Hong JS, et al. Metabolic tumor volume by positron emission tomography/computed tomography as a clinical parameter to determine therapeutic modality for early stage Hodgkin's lymphoma. Cancer Sci. 2013;104:1656–61.
21. Procházka V, Gawande RS, Cayci Z, Froelich JW, Cao Q, Wilke C, et al. Positron emission tomography-based assessment of metabolic tumor volume predicts survival after autologous hematopoietic cell transplantation for Hodgkin lymphoma. Biol Blood Marrow Transpl. 2018;24:64–70.
22. Plattel WJ, van den Berg A, Visser L, van der Graaf AM, Pruim J, Vos H, et al. Plasma thymus and activation-regulated chemokine as an early response marker in classical Hodgkin's lymphoma. Haematologica. 2012;97:410–5.
23. Plattel WJ, Visser L, Diepstra A, Glaudemans A, Nijland M, van Meerten T, et al. Interim thymus and activation regulated chemokine versus interim (18) F-fluorodeoxyglucose positron-emission tomography in classical Hodgkin lymphoma response evaluation. Br J Haematol. 2020;190:40–4.
24. van den Berg A, Visser L, Poppema S. High expression of the CC chemokine TARC in Reed-sternberg cells. Am J Pathol. 1999;154:1685–91.
25. Plattel WJ, Alsada ZN, van Imhoff GW, Diepstra A, van den Berg A, Visser L. Biomarkers for evaluation of treatment response in classical Hodgkin lymphoma: comparison of sGalectin-1, sCD163 and sCD30 with TARC. Br J Haematol. 2016;175:868–75.
26. Qin JQ, Yin H, Wu JZ, Chen RZ, Xia Y, Wang L, et al. 25-Hydroxy vitamin D deficiency predicts inferior prognosis in Hodgkin lymphoma. Leuk Res. 2021;105:106580.
27. Borchmann S, Cirillo M, Goergen H, Meder L, Sasse S, Kreissl S, et al. Pretreatment vitamin D deficiency is associated with impaired progression-free and overall survival in Hodgkin lymphoma. J Clin Oncol. 2019;37:3528–37.
28. Boellaard R, Delgado-Bolton R, Oyen WJ, Giammarile F, Tatsch K, Eschner W, et al. FDG PET/CT: EANM procedure guidelines for tumour imaging: version 2.0. Eur J Nucl Med Mol Imaging. 2015;42:328–54.
29. Kaalep A, Sera T, Oyen W, Krause BJ, Chiti A, Liu Y, et al. EANM/EARL FDG-PET/CT accreditation—summary results from the first 200 accredited imaging systems. Eur J Nucl Med Mol Imaging. 2018;45:412–22.
30. Boellaard R. Quantitative oncology molecular analysis suite: ACCURATE. J Nucl Med. 2018;59:1753.
31. Driessen J, Zwezerijnen GJ, Schöder H, Drees EE, Kersten MJ, Moskowitz AJ, et al. The impact of semi-automatic segmentation methods on metabolic tumor volume, intensity and dissemination radiomics in (18)F-FDG PET scans of patients with classical Hodgkin lymphoma. J Nucl Med. 2022;63:1424–30.
32. Eertink JJ, van de Brug T, Wiegers SE, Zwezerijnen GJC, Pfaehler EAG, Lugtenburg PJ, et al. 18F-FDG PET baseline radiomics features improve the prediction of treatment outcome in diffuse large B-cell lymphoma. Eur J Nucl Med Mol Imaging. 2021;49:932–43.
33. Barrington SF, Kluge R. FDG PET for therapy monitoring in Hodgkin and non-Hodgkin lymphomas. Eur J Nucl Med Mol Imaging. 2017;44:97–110.
34. Boktor RR, Walker G, Stacey R, Gledhill S, Pitman AG. Reference range for intrapatient variability in blood-pool and liver SUV for 18F-FDG PET. J Nucl Med. 2013;54:677–82.
35. Cheson BD, Fisher RI, Barrington SF, Cavalli F, Schwartz LH, Zucca E, et al. Recommendations for initial evaluation, staging, and response assessment of Hodgkin and non-Hodgkin lymphoma: the Lugano classification. J Clin Oncol. 2014;32:3059–68.
36. OpenClinica. LLC and collaborators; 2004–2018.
37. Josting A, Muller H, Borchmann P, Baars JW, Metzner B, Dohner H, et al. Dose intensity of chemotherapy in patients with relapsed Hodgkin's lymphoma. J Clin Oncol. 2010;28:5074–80.
38. von Tresckow B, Muller H, Eichenauer DA, Glossmann JP, Josting A, Boll B, et al. Outcome and risk factors of patients with Hodgkin Lymphoma who relapse or progress after autologous stem cell transplant. Leuk Lymphoma. 2014;55:1922–4.
39. Santoro A, Magagnoli M, Spina F, Pinotti G, Siracusano L, Michieli M, et al. Ifosfamide, gemcitabine, and vinorelbine: a new induction regimen for refractory and relapsed Hodgkin's lymphoma. Haematologica. 2007;92:35–41.
40. Chen R, Zinzani PL, Fanale MA, Armand P, Johnson NA, Brice P, et al. Phase II study of the efficacy and safety of pembrolizumab for relapsed/refractory classic Hodgkin lymphoma. J Clin Oncol. 2017;35:2125–32.
41. Armand P, Chen YB, Redd RA, Joyce RM, Bsat J, Jeter E, et al. PD-1 blockade with pembrolizumab for classical Hodgkin lymphoma after autologous stem cell transplantation. Blood. 2019;134:22–9.
42. Herrera AF, Chen L, Nieto Y, Holmberg L, Johnston PB, Mei M, et al. Consolidation with nivolumab and brentuximab Vedotin after autologous hematopoietic cell transplantation in patients with high-risk Hodgkin lymphoma. Blood. 2020;136:19–20.
43. Farina L, Rezzonico F, Spina F, Dodero A, Mazzocchi A, Crippa F, et al. Serum thymus and activation-regulated chemokine level monitoring may predict disease relapse detected by PET scan after reduced-intensity allogeneic stem cell transplantation in patients with Hodgkin lymphoma. Biol Blood Marrow Transpl. 2014;20:1982–8.

44. Sobesky S, Mammadova L, Cirillo M, Drees EEE, Mattlener J, Dörr H, et al. In-depth cell-free DNA sequencing reveals genomic landscape of Hodgkin's lymphoma and facilitates ultrasensitive residual disease detection. Med. 2021;2:1171–93.e11.
45. Ma Y, Visser L, Roelofsen H, de Vries M, Diepstra A, van Imhoff G, et al. Proteomics analysis of Hodgkin lymphoma: identification of new players involved in the cross-talk between HRS cells and infiltrating lymphocytes. Blood. 2008;111:2339–46.

ACKNOWLEDGEMENTS

We would like to dedicate this article to the memory of Professor Anton Hagenbeek, who together with MJK initiated this study. The authors would like to thank all patients who participated in the trial, the Transplant BRaVE-trial team of the Trial Office of the Amsterdam UMC, location AMC for their efforts in trial management and central data management, and the members of the Data Safety and Monitoring Board. The authors thank Marjolein Spiering, Edith van Dijkman, the data managers, trial nurses, lab- and pharmacy personnel for their essential assistance with collecting and managing the study data. The authors thank Nathalie Hijmering, HOVON Pathology Facility and Biobank, for biopsy collection and support of central pathology review.

AUTHOR CONTRIBUTIONS

MJK and Anton Hagenbeek designed and supervised the clinical study. AD supervised the biomarker study. All authors collected the data. JD and LV performed biomarker analysis. JD performed the PET segmentation. GJCZ and RB supervised the PET segmentation. DdJ and AD performed the central pathology review. AA, RV and GJCZ performed the central PET-CT review. JD performed the statistical analysis. JD wrote the manuscript with contributions from all authors. All authors interpreted the data, read, commented on, and approved the final version of the manuscript.

FUNDING

This work was supported by research funding from Takeda.

COMPETING INTERESTS

The study drug (BV) was provided for the study and the study was funded by Takeda. Takeda did not have any influence on the data analysis or the interpretation of the results. MJK: honoraria from and consulting/advisory role for BMS/Celgene, Kite, a Gilead Company, Miltenyi Biotech, Novartis, Adicet Bio and Roche; research funding from Kite, a Gilead Company, and Takeda; and travel support from Kite, A Gilead Company, Miltenyi Biotech, Novartis, and Roche. MH: Consultant/advisor: Roche, Takeda, Celgene, Genmab; Research support: Roche, Takeda, Celgene, Genmab, Novartis, Janssen, Incyte, Genentech. PJL: honoraria from and consulting/advisory role for Takeda, Servier, Roche, Genmab, AbbVie, Incyte, Regeneron, Celgene; Research funding from Takeda, Servier and Roche. FM: advisory boards for Roche, BMS, Genmab, Abbvie, Miltenyi, Novartis, Gilead, Asrtra Zeneca. Scientific lectures for Roche, Janssen. Consultancy for Roche, Genmab, Abbvie, Gilead. AD: Millennium/Takeda: Consultancy, Honoraria, Research Funding. TG: Millennium/Takeda: Honoraria, Gilead, Roche, MSD. JMZ: Consultant/advisor: Gilead, Roche, Takeda; Honoraria: Gilead, Roche, Takeda, Janssen. DdJ: Consultant/advisor: Takeda. SHT: Consultant/advisor: Takeda, Kite/Gilead; Research Funding: Beigene. The other authors declare no potential conflicts of interest.

ADDITIONAL INFORMATION

Supplementary information The online version contains supplementary material available at https://doi.org/10.1038/s41375-022-01717-8.

Correspondence and requests for materials should be addressed to Marie José. Kersten or Arjan Diepstra.

Reprints and permission information is available at http://www.nature.com/reprints

Publisher's note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

ON BEHALF OF THE HOVON LUNENBURG LYMPHOMA PHASE I/II CONSORTIUM (LLPC)

Marie José Kersten[1,15]✉, Sanne H. Tonino[1], Josée M. Zijlstra[3], Pieternella J. Lugtenburg[4] and Marcel Nijland[9]